INFLUENCE OF COMPOSITION OF PLANT MATERIALS ON PROPERTIES OF THE DECOMPOSED RESIDUES[1]

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The origin, composition, and properties of soil organic matter have long been subjects of much study and controversy. The term "humus," which has been highly abused in designating different organic substances, dates back to the time of the Romans, when it was frequently used to designate the soil as a whole (27). Wallerius (28), in 1761, was the first to define "humus" in terms of decomposed organic matter, but the prevailing ideas concerning the chemical nature of organic matter and the mechanism of its formation were still very vague.

Even in the early part of the present century many still believed that the "humic acids" originated from carbohydrates, regardless of whether they were natural compounds or preparations obtained by treatment of carbohydrates with acids (4, 8, 11, 27). Eller helped to clear up some of the confusion in 1925 when he found that on oxidation of phenols and their derivatives, products were obtained which were similar in their chemical and physical properties to the natural "humic acids" (5). According to him (6), the "humic acids" obtained by treatment of carbohydrates with mineral acids have nothing in common with the natural products, in spite of a superficial similarity. Waksman has said that one may feel justified in abandoning without reservation the whole nomenclature of humic acids with the possible exception, because of its historical importance, of the term "humus"; and that this term should be used to designate the organic matter of the soil as a whole (27).

The role of lignin in the formation of "humus" has been investigated by many workers, and though their conclusions have varied greatly, it is now accepted that lignin plays an important part in the formation of "humus" and in its chemical properties. Some of the conflicting views have been caused by the difficulties in estimating the exact amount of lignin present in decomposing plant materials.

Much effort has been spent in trying to maintain an adequate supply of organic matter in agricultural soils. Because organic matter is a constituent which should be maintained in the soil in relatively large amounts and at the same time one which must be continuously in process of decomposition, the maintenance of a good supply in cultivated soils will inevitably involve periodic additions.

In view of the fact that the nature and composition of soil organic matter depend on the nature of the parent material from which the organic matter is derived, and through their influences on the actual population, the conditions under which that material is decomposed, more advantageous additions might be made if more information were available on the amount of organic matter derived from different plants or the constituents thereof, and on the effect of this material on the properties of the soil. The purposes of this work were (*a*) to ascertain the effect of varying the proportions of certain major constituents of plant materials on the course of decomposition and (*b*) to determine the contribution made by individual plant constituents to the properties of the decomposed residues obtained therefrom and the effects of the residues on some properties of soil.

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Oxidation studies were planned in the hope of developing a method that would facilitate the study of the nature of organic matter in soils and determine the quantities therein.

EXPERIMENTAL PROCEDURE

Materials used

Thurman loamy fine sand was chosen for this work because of its low organic matter content, about 1 per cent. In such a soil, the effects produced by the added organic materials should be most pronounced and probably most easily measured.

The organic materials added were untreated oat straw; oat straw preparations with varying proportions of lignin, cellulose, and hemicellulose; dextrose; soybean stalks; and alfalfa. A partial analysis of these materials is given in table 1, and the methods of preparations of the oat straw materials follow.

Pretreatment of oat straw

Inasmuch as it was intended to study the decomposition of the treated and the untreated straw both in the absence and in the presence of soil, and because finely ground straw could not be used in the former circumstance, the treatments were applied to straw that had been cut into short lengths in an ensilage cutter. That portion of the preparation required for addition to soil was later ground more finely in a Wiley mill.

Sulfuric acid extraction of straw. The chief purpose of this treatment was to remove the hemicelluloses and other more hydrolyzable constituents, and to leave a residue consisting primarily of cellulose and lignin. It was carried out by boiling 150 gm. of straw in 5 liters of 5 per cent sulfuric acid for 1 hour, then washing until free of acid. The loss on hydrolysis was about 40 per cent. The lignin content of the residue was 26 per cent, an increase of 53 per cent over that of the original straw.

Chlorine, alcoholic-ethanolamine extraction of straw (23). The purpose of this treatment was to accomplish partial delignification without concurrent removal of hemicellulose or other non-water-soluble polysaccharides.

Water was added to 250 gm. of straw and allowed to remain until the straw was wet. Then 400 ml. of sodium hypochlorite (5.25 per cent Cl) was added, and the solution was made just acid with sulfuric acid. After thorough stirring, this was allowed to stand for 15 minutes, then the solution was drained off. Sufficient hot 3 per cent alcoholic enthanolamine (3 parts of monoethanolamine added to 97 parts of 95 per cent alcohol) was added completely to cover the material, and stirred thoroughly. This was allowed to stand 5 minutes and was then drained off. The residue was washed with cold distilled water until neutral to litmus.

This treatment brought about incomplete delignification, the content in the residue being only 6 per cent less than that of the original straw. Though it would be anticipated that the nitrogen content of the straw would be reduced by the treatment, inasmuch as proteins would be attacked by the chlorination, the apparent nitrogen content was in fact increased, probably as a result of condensation of some ethanolamine with the residual lignin. Other constituents soluble in water and alcohol were removed, and the total loss was 25 per cent of the original.

TABLE 1

Composition of materials used in the decomposition studies

MATERIAL	LOSS BY TREATMENT	NITROGEN	CARBON	LIGNIN	LIGNIN REMOVED BY TREATMENT	CELLULOSD*	FURFURALDEHYDE FROM CELLULOSE*	XYLAN IN CELLULOSE	TOTAL FURFURALDEHYDE*	POLYURONIDE HEMICELLULOSE FURFURALDEHYDE
	%	%	%	%	%	%	%	%	%	%
Oat straw untreated..	..	0.39	43.62	17.1	....	49.6	8.2	12.7	15.6	7.4
5% H_2SO_4-extracted oat straw...........	40	0.25	47.01	26.3	7.9	63.0	6.0	9.4	8.6	2.6
Cl-amine-extracted oat straw...........	26	0.45	46.01	16.0	30.8	62.2	9.1	14.0	17.4	8.3
0.5% Na_2CO_3-extracted oat straw....	29	0.27	45.93	17.2	24.7	61.8	9.5	14.7	18.3	8.8
0.5% NaOH-extracted oat straw...........	40	0.09	45.92	10.3	64.0	73.5	11.9	18.4	16.7	4.8
NaOH, NaOCl, Na_2SO_3-extracted oat straw (cellulose)....	60	0.07	43.08	3.4	91.9	91.5	14.6	22.5	16.6	2.0
Thurman fine sandy loam..............	..	0.05	0.62	...	....	....	....	....	....	...
Dextrose.............	..	....	40.00	...	....	....	....	....	....	...
Soybean stalks.......	..	0.76	46.85	15.6	....	....	....	....	....	...
Alfalfa..............	..	3.54	45.70	15.8	....	....	....	....	....	...

* Analyses by L. N. Spohnheimer.

Sodium carbonate (0.5 per cent) extraction of straw. This extraction was applied to accomplish extensive removal of the hemicelluloses and partial delignification.

About 150 gm. of straw was boiled in 5 liters of 0.5 per cent sodium carbonate solution for 15 minutes; then the residue was washed free of carbonate with cold distilled water. The total loss was 29 per cent.

Sodium hydroxide (0.5 per cent) extraction of straw. The purpose of this treatment, which was carried out by boiling 150 gm. of straw in 5 liters of 0.5 per cent sodium hydroxide for 30 minutes, was to bring about more extensive delignification than that accomplished by the carbonate. The effect on the hemicelluloses and nitrogenous constituents would not be greatly different from that in foregoing extraction. The loss on extraction was 40 per cent, and the lignin content of the residue was only 60 per cent as high as in the original straw. The nitrogen content was only about one-fourth as high.

"Cellulose" preparation (15). The purpose of this treatment was to remove virtually all incrusting materials from the cell walls and to leave only that portion of the cellulose resistant to extraction by alkali. The total loss caused by the treatment was 60 per cent.

The treatments consisted first of boiling 150 gm. of straw in 5 liters of 1 per cent sodium hydroxide for 10 minutes, and washing free of hydroxide with water. The straw was then covered with water, 250 ml. of sodium hypochlorite (0.5 per cent Cl) was added, the mixture was made acid with sulfuric acid and stirred well. After 15 minutes the sodium hypochlorite and sulfuric acid treatments were repeated, the mixture was allowed to stand another 15 minutes, and was drained off.

The residue was then boiled with 3 per cent sodium sulfite for 20 minutes, drained off, and washed with distilled water until neutral. The structure of the straw was not maintained in this preparation.

Soil treatments

Twenty pots, each containing 2 kilos of Thurman soil, were set up with the addition of 10 per cent finely ground organic material in the form of oat straw, the oat straw residues from the various straw treatments, soybean stalks, alfalfa, and dextrose. All were made up to a C:N ratio of 29:1 with ammonium nitrate, except one set of soybean stalks which was left at the original ratio of 62:1, and the alfalfa which was left at 13:1. Two pots of soil were set up as checks. To each was added 6 gm. of calcium carbonate. To the pots receiving the plant residues, 900 ml. of water was added; to those receiving dextrose, 315 ml.; and to the check soils, 290 ml. Water was added regularly as needed.

Decomposition of straw separate from soil

The oat straw residues, except the "cellulose" preparation, were placed in fruit jars. Fifty grams of each material was put in a jar and made up to a C:N ratio of 29:1 with ammonium nitrate, and 175 ml. of water added. The jars, set up in triplicate, were inoculated with an extract made from the original oat straw and a soil high in organic matter.

Sampling and analyses

Each pot of soil was mixed thoroughly and sampled 105, 328, 580, and 833 days after the experiment was started. The samples were analyzed at each period in order to follow the changes taking place during decomposition.

One jar of each plant material was analyzed after 112 days; one jar after 322 days; and the last after 596 days.

In the analysis of the soil and organic materials, the official method of the A.O.A.C. (1) was used for determination of total nitrogen. Total carbon was determined by the dry combustion method (30). Base-exchange capacity was determined by the barium acetate, calcium acetate method of Millar, Smith, and Brown (13). Organic exchange capacity was derived by oxidizing the soil with H_2O_2, determining inorganic exchange capacity, then subtracting this from the total exchange capacity. Lignin was determined by the method of Norman and Jenkins (16). The hypoiodite oxidations of the soils and plant materials followed a method previously reported from this laboratory (18). The hypochlorite oxidations were according to a method reported by Norman (17).

EXPERIMENTAL RESULTS AND DISCUSSION

Effects of composition of plant residues on amount and nature of soil organic matter

All organic materials added to the soil in this study were at first rapidly decomposed, and after 105 days from 50 to 95 per cent of the carbon added had disappeared (table 2). Except where dextrose was added, there was considerable decomposition after 105 days, but there was little after 328 days regardless of the nature of the organic material added. The oat straw "cellulose" preparation, which contained 3.4 per cent lignin, was much more rapidly and completely decomposed than any of the other additions of plant materials. After 833 days' decomposition, only 7.4 per cent of the "cellulose" preparation remained in the soil, whereas 22.6 per cent of the acid-hydrolyzed straw was still present. This latter material was the most resistant to decomposition, no doubt because of its large lignin content and the extensive removal of hemicelluloses and other more soluble constituents. The degree of decomposition of the other oat straw materials was intermediate between the "cellulose" and the acid-hydrolyzed straw, depending on the lignin content of the material and the degree of removal of cellulose and other materials by the pretreatments.

To study the nature of the residues from these materials, the soils were treated with 72 per cent sulfuric acid as in the procedure used to determine lignin, and the content of acid-resistant organic material was then determined in the soils (table 3). Wide variations were found in the percentage of acid-resistant material, but even greater variations were found in the quantities of acid-resistant material as compared to that in check soil. The total amount present in the soil which received acid-hydrolyzed straw was almost three times as great as that in the check soil and much higher than where any other material was used. It is also of interest to note that where dextrose was added, the amount of acid-resistantant material was less than in the check soil, even though the total carbon present was higher in the former. This is indicative of the fact that the synthesized tissues produced under these circumstances contain little if any acid-resistant constituents. When the "cellulose" preparation was added, the increase was only about one-fourth as great as when the acid-hydrolyzed straw was added.

The quantities of nitrogen resistant to removal by this strong acid treatment in these soils were higher than in the check soil, but removal of nitrogen by this treatment was generally more extensive than that of carbon. In all instances the C:N ratios were higher after the strong acid treatment.

TABLE 2

Changes in organic carbon in soil during decomposition

ORGANIC MATERIAL ADDED TO SOIL	CARBON CONTENT				PER CENT OF ADDED CARBON LEFT AFTER 833 DAYS
	After 105 days	After 328 days	After 580 days	After 833 days	
	%	%	%	%	
Check	0.67	0.66	0.61	0.59	....
Oat straw untreated	2.04	1.36	1.21	1.13	12.4
5% H_2SO_4-hydrolyzed straw	2.65	1.72	1.72	1.65	22.6
Cl-amine-extracted straw	2.75	1.52	1.48	1.41	17.8
0.5% Na_2CO_3-extracted straw	2.32	1.44	1.32	1.29	15.2
0.5%-NaOH extracted straw	2.25	1.35	1.21	1.17	12.6
"Cellulose" from oat straw	1.46	1.23	1.11	0.91	7.4
Dextrose	0.83	0.76	0.71	0.70	2.8
Alfalfa	2.30	1.41	1.34	1.30	15.6
Soybeans 62/1 C:N	2.91	1.33	1.27	1.22	13.5
Soybeans 29/1 C:N	3.35	1.35	1.34	1.22	13.5

TABLE 3

Effect of 72 per cent H_2SO_4 on carbon and nitrogen contents of soil after decomposition for 833 days

ORGANIC MATTER ADDED TO SOIL	CARBON		NITROGEN		C/N RATIOS		RESISTANT MATERIAL IN SOIL ORGANIC MATTER	INCREASE IN ACID-RESISTANT MATERIAL IN SOIL
	Before acid treatment	After acid treatment	Before acid treatment	After acid treatment	Before acid treatment	After acid treatment		
	%	%	%	%			%	%
Check	0.58	0.39	0.067	0.034	8.7	11.5	67	....
Oat straw untreated	1.19	0.71	0.119	0.066	10.0	10.8	60	82
5% H_2SO_4-hydrolyzed straw	1.50	1.15	0.145	0.073	10.3	15.8	72	194
Cl-amine-extracted straw	1.46	0.84	0.132	0.069	11.1	12.2	58	115
0.5% Na_2CO_3-extracted straw	1.34	0.89	0.127	0.074	10.6	12.0	66	128
0.5% NaOH-extracted straw	1.15	0.80	0.131	0.057	8.8	14.0	70	106
"Cellulose" from oat straw	0.93	0.59	0.101	0.055	9.2	10.7	63	51
Dextrose	0.71	0.38	0.103	0.043	7.9	8.8	54	−2.5
Alfalfa	1.29	0.82	0.156	0.094	8.3	8.7	64	110
Soybean 62/1 C:N	1.21	0.71	0.123	0.070	9.8	10.1	59	82
Soybean 29/1 C:N	1.26	0.70	0.135	0.069	9.3	10.1	56	80

These differences in the residues can be attributed only to differences in composition of the organic materials added. Some fraction of these materials was less susceptible to decomposition than others. Since lignin was the constituent that varied most widely in amount and since the materials that were at least extensively decomposed were those that were highest in lignin, it is deduced that lignin was largely responsible for this resistance to decomposition and for the increased amount of acid-resistant material present in the residues.

The decomposition of these plant materials in the absence of soil also showed that those high in lignin were more resistant to decomposition by microorganisms (table 4). These materials with a high lignin content were decomposed much more thoroughly when incorporated in soil. This is most likely due to the population differences in the two environments. Organisms that can perhaps bring about a slow attack on lignified tissues in the soil may be unadapted to the highly aerobic, drier conditions in the jars. The amount of apparent lignin remaining in these materials after 596 days of decomposition also helps to emphasize the resistance of this constituent to destruction by microorganisms (table 5). Even though the untreated straw was 77 per cent decomposed at this time, only about half of the lignin therein was destroyed and only a small amount of the lignin in any other material was decomposed. The apparent lignin content of the Cl-amine-treated straw increased slightly, and that of the sodium-hydroxide-extracted straw increased considerably. The increase in the latter material is attributed to synthesized products formed during the decomposition process. In the Cl-

TABLE 4

Decomposition of plant materials in absence of soil

PLANT MATERIAL	DECOMPOSITION		
	After 112 days	After 322 days	After 596 days
	%	%	%
Oat straw untreated	47.4	73.8	77.2
5% H_2SO_4-hydrolyzed straw	5.3	29.4	18.4
Cl-amine-extracted straw	8.0	28.0	31.9
0.5% Na_2CO_4-extracted straw	19.9	41.6	52.0
0.5% NaOH-extracted straw	12.4	46.4	77.8

TABLE 5

Apparent lignin content of plant materials during decomposition in absence of soil

PLANT MATERIAL	APPARENT LIGNIN CONTENT			
	At beginning	After 112 days	After 322 days	After 596 days
	gm.*	gm.*	gm.*	gm.*
Oat straw untreated	17.1	15.0	9.3	8.2
5% H_2SO_4-hydrolyzed straw	26.3	25.3	25.3	22.5
Cl-amine-extracted straw	16.0	17.5	17.6	17.2
0.5% Na CO_4-extracted straw	17.2	17.7	17.9	15.2
0.5% NaOH-extracted straw	10.3	11.6	13.7	14.1

* Per 100 gm. original material.

amine-treated straw, this increase may be due in part to the effects of the amine treatment. There is an indication in the increase in the nitrogen content of the residue after the Cl-amine treatment (table 1) that some combination between amine and lignin may have taken place.

It has been reported by other workers that lignin is of great importance in soil organic matter. Fischer (7) demonstrated that in the formation of peat the celluloses of plant residues change to CO_2 and water, whereas the lignin accumulates, even though it may be altered somewhat. In the work reported here, it was found, with one exception, that part of the lignin was decomposed but not to the same degree as cellulose and other constituents. Wehmer (29) also reported that lignin was attacked less than other constituents and believed that it was converted into humic acids. Similar results have been reported by Rose and Lisse (20) and others (3, 24, 25).

Even though decomposition of the oat straw preparations was slow after 833 days, it could not be considered to have stopped completely, and part of the lignin was definitely decomposed. Several workers (12, 21, 22) have reported rather large losses of lignin over periods of several months, and Phillips (19) has concluded that, under proper conditions, soil organisms are capable of decomposing lignin in plant materials and, under suitable conditions, the rate of decomposition may be as great as that of cellulose and pentosans. This latter statement was not found to be true for the extracted lignified materials in this study.

Exchange studies

Further information on the nature of the residues from these materials was brought out by studies on total exchange and organic exchange capacities (table 6). The total exchange capacities were highest in the soils receiving the plant materials high in lignin, but the results are more striking when the increases in organic exchange capacities are considered. At the end of the experiment the soils receiving the materials high in lignin had organic exchange capacities almost three times as great as that of the check soil, whereas the soil receiving the "cellulose" preparations had an increase only one-fourth as great. However, increases in exchange capacities, like other properties, were not proportional to increases in lignin content of the original material, partly because of differences in the nature of the decomposition and the residues, caused by pretreatment of the straw.

These relatively large increases in exchange capacities are believed to be caused largely by the lignin. According to Mitchell (14), the "lignin-humus" fraction is the most important fraction of soil organic matter possessing exchange properties, since it appears to give the organic matter its more or less permanent exchange properties. McGeorge (9, 10) has said that the exchange property of organic matter is, for the most part, due to lignin compounds. Bartlett and Norman (2) found that lignin isolated from residues of different plants possessed very different exchange properties, and after decomposition for 6 months, the exchange capacities of the lignin fractions were found to have been increased severalfold. This indicates that lignin in soil organic matter becomes more active after decomposition has taken place.

The exchange capacities of the straw preparations decomposed in the absence of soil (table 7) were variable. Here again, they were not directly related to the lignin or apparent lignin contents. The more extensively decomposed materials had by far the greatest capacities per unit of residue. The differences are not so great, when the exchange capacities are calculated on the basis of 100 gm. of the original material (table 7). It was found, on this basis, that all except one of the materials had decreased exchange capacities after 596 days. Therefore, even though the residues were more active, it is evident that some material having exchange properties was being decomposed, and faster than the increase in the activity of the residues.

TABLE 6

Exchange capacity of soil and of organic matter during decomposition

(m.e. Ba per 100 gm. soil)

ORGANIC MATERIAL ADDED TO SOIL	TOTAL EXCHANGE CAPACITY OF SOIL				ORGANIC EXCHANGE CAPACITY OF SOIL			
	After 105 days	After 328 days	After 580 days	After 833 days	After 105 days	After 328 days	After 580 days	After 833 days
Check	8.0	7.9	8.3	8.3	2.1	2.0	2.3	2.3
Oat straw untreated	11.3	10.9	13.3	12.8	5.3	5.0	7.4	6.8
5% H_2SO_4-hydrolyzed straw	11.1	11.5	12.3	12.3	5.2	5.5	6.3	6.4
Cl-amine-extracted straw	10.7	12.7	12.2	11.2	4.7	6.7	6.2	5.3
0.5% Na_2CO_3-extracted straw	11.4	12.1	13.1	12.0	5.4	6.2	7.1	6.0
0.5% NaOH-extracted straw	9.4	11.2	11.1	10.3	3.4	5.3	5.1	4.3
"Cellulose" from oat straw	9.2	10.6	9.9	9.6	3.3	4.6	3.9	3.6
Dextrose	8.4	8.7	9.3	9.8	2.4	2.7	3.4	3.8
Alfalfa	10.0	12.6	12.9	12.8	4.1	6.6	7.0	6.8
Soybeans 62/1 C:N	11.4	11.6	11.8	11.7	5.4	5.8	5.8	5.7
Soybeans 29/1 C:N	11.2	12.1	11.7	11.7	5.3	6.1	5.8	5.8

TABLE 7

Base exchange capacity of plant materials during decomposition in absence of soil

PLANT MATERIAL	ORIGINAL MATERIAL (M.E. Ba PER 100 GM.)				RESIDUAL MATERIAL (M.E. PER 100 GM.)		
	At beginning	After 112 days	After 322 days	After 596 days	After 112 days	After 322 days	After 596 days
Oat straw untreated	42.3	60.1	58.8	51.4	114.3	224.8	225.0
5% H_2SO_4-hydrolyzed straw	34.8	27.9	26.4	29.0	29.5	37.4	35.5
Cl-amine-extracted straw	34.3	28.1	23.8	25.0	30.6	33.0	36.7
0.5% Na_2CO_3-extracted straw	42.3	41.9	37.7	38.7	52.3	64.6	80.8
0.5% NaOH-extracted straw	32.9	34.5	22.8	23.1	39.5	42.5	104.0

Nitrogen relationships

As the decomposition of the plant materials proceeded and the carbon content decreased, the amount of nitrogen in the soils in all treatments also decreased materially (table 8), though not to the same extent as the carbon. The soil receiving the dextrose showed the greatest decrease in nitrogen, most of which occurred in 105 days, whereas the soil receiving the acid-hydrolyzed straw showed the smallest decrease. The soils contained only relatively small amounts of nitrate nitrogen or ammonia nitrogen at any time and since there were no losses from leaching and none from plant removal, the only loss was into the atmosphere.

Because of this decrease in nitrogen content as decomposition proceeded, the conclusion might be drawn that nitrogen was conserved only because of the resistance of the different materials to decomposition. Upon closer observation, however, other factors influencing the conservation of nitrogen may be pointed out. The nitrogen contents decreased materially, as evidenced by the C:N ratio, but not so rapidly as the materials decomposed (table 9). It may be concluded, therefore, that nitrogen was conserved by the partly decomposed residues, probably as microbial tissue. If this were not true, the C:N ratios would have been much wider as decomposition proceeded. After 833 days the ratios varied from 8.8:1 to 11.8:1, the latter being in the soil to which acid-hydrolyzed straw had been added. This soil also had a higher percentage of total nitrogen than any of the others receiving oat straw preparations.

TABLE 8

Total nitrogen in soil during decomposition

ORGANIC MATERIAL ADDED TO SOIL	NITROGEN CONTENT			
	After 105 days	After 328 days	After 580 days	After 833 days
	%	%	%	%
Check	0.067	0.058	0.058	0.067
Oat straw untreated	0.172	0.146	0.134	0.119
5% H_2SO_4-hydrolyzed straw	0.167	0.161	0.147	0.140
Cl-amine-extracted straw	0.161	0.147	0.142	0.130
0.5% Na_2CO_3-extracted straw	0.173	0.152	0.136	0.130
0.5% NaOH-extracted straw	0.152	0.128	0.118	0.132
"Cellulose" from oat straw	0.144	0.112	0.104	0.099
Dextrose	0.106	0.092	0.097	0.090
Alfalfa	0.240	0.202	0.176	0.160
Soybeans 62/1 C:N	0.136	0.158	0.133	0.122
Soybeans 29/1 C:N	0.159	0.168	0.130	0.134

Waksman and Hutchings (26) reported that lignin is important in nitrogen conservation, acting as a buffer for the adsorption of ammonia. This was not true in this study, for but little ammonia was present at any time. They said also that lignin "fixes" protein in the form of lignoprotein complexes and thus helps to keep the nitrogen content of soil at a higher level. This seems to have been true in this work, for soils receiving materials high in lignin had much greater nitrogen supplies finally than did those receiving dextrose, "cellulose," or other oat straw preparations low in lignin, although the nitrogen levels of all were identical initially. The nitrogen conservation, however, was not proportionate to the lignin content, probably because increased activity of the lignin in the highly decomposed materials formed more ligno-protein complexes. Large losses of nitrogen occurred when the materials were decomposed in the absence of soil (table 10), though the nitrogen content of some of the residues after 596 days was high, particularly in the untreated straw, which was extensively decomposed.

TABLE 9

Carbon: nitrogen ratios of soil during decomposition

ORGANIC MATERIAL ADDED TO SOIL	CARBON: NITROGEN RATIO			
	After 105 days	After 328 days	After 580 days	After 833 days
Check	10.1	11.1	10.4	8.8
Oat straw untreated	11.8	9.3	9.0	9.5
5% H_2SO_4-hydrolyzed straw	15.9	10.7	11.7	11.8
Cl-amine-extracted straw	17.1	10.4	10.4	10.9
0.5% Na_2CO_3-extracted straw	14.0	9.2	9.7	10.0
0.5% NaOH-extracted straw	14.8	10.5	10.3	8.8
"Cellulose" from oat straw	10.1	11.0	10.7	9.7
Dextrose	7.8	8.4	7.3	7.9
Alfalfa	9.6	7.0	7.6	8.1
Soybeans 62/1 C:N	21.4	8.4	9.5	10.0
Soybeans 29/1 C:N	21.1	8.1	10.3	9.0

TABLE 10

Changes in total nitrogen content during decomposition of plant materials in absence of soil

PLANT MATERIAL	NITROGEN CONTENT			ORIGINAL N ACCOUNTED FOR AFTER 596 DAYS*
	After 112 days	After 322 days	After 596 days	
	%	%	%	%
Oat straw untreated	1.97	2.80	3.82	55
5% H_2SO_4-hydrolyzed straw	1.25	1.45	1.12	56
Cl-amine-extracted straw	1.18	1.29	1.36	49
0.5% Na_2CO_3-extracted straw	1.28	1.54	1.82	54
0.5% NaOH-extracted straw	1.10	1.68	2.02	27

* Original N = N in plant material plus N added as NH_4NO_3.

The lignin contents of the chemically extracted straw preparations were higher than in the untreated straw, but the protein content was higher in the latter because of greater microbial synthesis and greater formation of lignoprotein complexes.

Oxidation studies

Application of hypochlorite and hypoiodite oxidative procedures to the treated soils and decomposed plant residues obtained in these investigations permits of an evaluation of the suitability of these methods in following changes in the composition of decomposing plant materials, or in comparing the organic matter of soils. The methods were devised on the grounds that changes in amounts of those constituents susceptible to oxidation could readily be followed.

The results of the oxidation of the soils and the straw materials show that these procedures may be used to throw light on the nature of the soil organic matter but not on the quantity, if there are variations in the nature of the material. This is true because certain constituents of organic matter are more susceptible than others to oxidation by these reagents. Lignin has been found to be the major plant constituent most susceptible to these oxidations. Protein is also oxidized by these reagents but to a lesser extent than lignin. Therefore, the extent of oxidation of organic matter by these reagents will be determined largely by the lignin and protein contents of the materials and by the extent and nature of their decomposition.

TABLE 11

Hypoiodite oxidation of soil during decomposition

(m.e. I_2 per 100 gm. soil)

ORGANIC MATERIAL ADDED TO SOIL	OXIDATION OF SOILS			
	After 105 days	After 328 days	After 580 days	After 833 days
Check	3.90	3.90	4.80	4.50
Oat straw untreated	15.35	8.85	10.10	11.65
5% H_2SO_4-hydrolyzed straw	25.50	16.05	20.00	19.55
Cl-amine-extracted straw	17.60	11.25	12.65	13.95
0.5% Na_2CO_3-extracted	19.45	9.85	10.85	13.80
0.5% NaOH-extracted straw	15.90	7.60	8.40	11.55
"Cellulose" from oat straw	15.20	8.40	8.00	9.55
Dextrose	13.65	6.20	6.35	6.75
Alfalfa	15.50	9.00	10.10	12.95
Soybeans 62/1 C:N	13.55	6.60	8.70	8.40
Soybeans 29/1 C:N	12.95	7.60	8.85	8.40

It was found that the organic matter in the soils receiving the high lignin materials was more susceptible to hypoiodite oxidation than was that in the siols receiving the materials low in lignin (table 11). The soil receiving the acid-treated straw was more than four times as active toward this reagent as the check soil. The carbon content was less than three times as great, showing that the organic matter in the soils receiving the acid-hydrolyzed straw was more oxidizable than that in the check soil, as shown by the I_2:C ratio in table 12. The same general relation was shown by oxidizing the soils that were treated with 72 per cent sulfuric acid (table 13). The I_2:C ratios for the materials with relatively high lignin contents were fairly constant and were higher than the ratios for the check soil and soils receiving materials low in lignin.

The results of the hypoiodite oxidation of the decomposed straw material (table 14) show that the residues were generally more highly oxidizable as decomposition proceeded and as the lignin content increased. The apparent lignin

TABLE 12

Iodine: carbon ratios of soil during decomposition

ORGANIC MATERIAL ADDED TO SOIL	M.E. I_2: TOTAL CARBON RATIO			
	After 105 days	After 328 days	After 580 days	After 833 days
Check	5.82	5.97	7.90	7.63
Oat straw untreated	7.57	6.54	8.41	10.37
5% H_2SO_4-hydrolyzed straw	9.94	9.37	11.62	11.84
Cl-amine-extracted straw	6.41	7.42	8.55	9.87
0.5% Na_2CO_3-extracted straw	8.48	6.84	8.20	10.64
0.5% NaOH-extracted straw	7.12	5.65	6.98	9.85
"Cellulose" from oat straw	10.46	6.83	7.25	10.50
Dextrose	16.50	8.16	8.97	9.04
Alfalfa	8.75	6.40	7.56	9.95
Soybeans 62/1 C:N	4.64	5.06	6.89	6.92
Soybeans 29/1 C:N	3.87	5.65	6.61	6.95

TABLE 13

Hypoiodite oxidation of 72 per cent sulfuric-acid extracted soil after decomposition for 833 days

ORGANIC MATTER ADDED TO SOIL	M.E. I_2 PER 100 GM. SOIL		M.E. I_2/C	
	Before acid treatment	After acid treatment	Before acid treatment	After acid treatment
Check	4.50	3.09	7.76	7.91
Oat straw untreated	11.81	8.16	9.92	11.5
5% H_2SO_4-hydrolyzed straw	19.69	12.38	13.13	10.74
Cl-amine-extracted straw	14.35	9.87	9.8	11.67
0.5% Na_2CO_3-extracted straw	13.05	9.56	10.07	10.74
0.5% NaOH-extracted straw	11.25	9.20	9.78	11.5
"Cellulose" from oat straw	10.12	5.14	10.88	8.71
Dextrose	6.47	3.09	9.11	8.13
Alfalfa	12.37	8.84	9.74	10.78
Soybeans 62/1 C:N	8.44	7.34	6.98	10.34
Soybeans 29/1 C:N	8.44	7.31	6.77	10.44

TABLE 14

Hypoiodite oxidation of straw during decomposition in absence of soil

(m.e. I per 100 gm.)

PLANT MATERIAL	RESIDUAL MATERIAL			ORIGINAL MATERIAL		
	After 112 days	After 322 days	After 596 days	After 112 days	After 322 days	After 596 days
Oat straw untreated	231	284	236	121.6	73.2	53.7
5% H_2SO_4-hydrolyzed straw	245	340	290	232.0	240.0	192.6
Cl-amine-extracted straw	155	160	197	142.6	115.2	134.1
0.5% Na_2CO_3-extracted straw	186	211	228	149.1	123.2	109.4
0.5% NaOH-extracted straw	183	211	318	160.3	113.0	70.5

content of the sodium-hydroxide-treated straw was very high at the end of the experiment, and this residue was very highly oxidizable by hypoiodite.

It seems, from these data, that the hypoiodite oxidation is closely related to the lignin content of the residues. When these residues are derived from the same source, variation in oxidation values will be due largely to differences in lignin content of the residues; but when the residues are derived from different materials, variations in oxidation values will probably be due to differences in the nature of the lignin as well as its content.

It is also of interest to note the amount of iodine utilized per 100 gm. soil before and after the strong acid treatment. In the soils receiving the materials high in lignin, the values after acid treatment were about two-thirds as large as before, whereas soils receiving dextrose and "cellulose" had values only one-half as large after acid treatment as before. This indicates that there was a difference in the nature of these residues, the former group probably being higher in lignin than the latter, and that present in the former group being more susceptible to oxidation. That the lignin was more active was brought out by the I_2:C ratios after acid treatment.

In previous work reported from this laboratory (18), wide variations in hypoiodite oxidation were found for the organic carbon in different soils, as well as for various depths in the same soil. These variations may be due to differences in the amount or nature of lignin, when two soils are studied, but when such a study is in the different layers of the same soil the variations are probably due to the greater percentage of lignin in the organic matter of the more active layer.

Bartlett and Norman (2) reported an average of 27 m. e. iodine utilized per gram of lignin carbon for a number of lignin preparations. The reactivity of the residues obtained in this work was lower than might have been expected on the basis of the known lignin contents, and as already suggested, there seems to be some form of combination between lignin and other constituents as a result of which the reactivity of the lignin component is reduced (18). Even after strong acid treatment, however, the material remaining in the soils did not approach the figures given for lignin directly isolated from undecomposed plant materials.

The degree of oxidation by hypoiodite is limited because of the nature of this reagent as an oxidant. When iodine is added to sodium hydroxide with the formation of hypoiodite, the reaction

$$3\ NaOI \rightarrow NaIO_3 + 2\ NaI$$

begins and proceeds rapidly at ordinary gemperatures so that at the end of 20 to 30 minutes no free hypoiodite remains. When an oxidizable material is treated with this reagent, the oxidation of the material will end when the foregoing reaction had ended, even though much of the material may still be susceptible to further oxidation.

The hypochlorite oxidation (tables 15 and 16) is far more extensive than that of the hypoiodite, chiefly because of the difference between the two reagents. In contrast to hypoiodite oxidation, hypochlorite oxidation is initially more active and may continue for several days though the rate is much reduced after 18 to 14 hours.

In this work, the hypochlorite oxidation seemed to be related more closely to the nitrogen than to the lignin content, but this was not definitely established. At the end of the experiment the oxidation values for the untreated straw and the NaOH-extracted straw were about the same, yet the former material was much higher in organic nitrogen than the latter. The latter, however, had a much greater apparent lignin content, and since lignin is also extensively oxidized by hypochlorite, it was probably this greater amount of lignin that caused the total oxidation value to be as large as that for the untreated straw. The oxidation of the other residues followed fairly closely the protein content, and here, too, the variation was probably caused, at least in part, by the variations in the amount and nature of the lignin.

TABLE 15

Hypochlorite oxidation of soils during decomposition

(m.e. Cl per 100 gm. soil)

ORGANIC MATERIAL ADDED TO SOIL	OXIDATION OF SOILS			
	After 103 days	After 328 days	After 580 days	After 833 days
Check	44.5	50.0	40.6	43.0
Oat straw untreated	198.0	129.5	132.5	168.5
5% H_2SO_4-hydrolyzed straw	248.0	190.0	158.7	168.5
Cl-amine-extracted straw	207.4	164.5	134.4	147.5
0.5% Na_2CO_3-extracted straw	188.0	146.5	133.3	151.4
0.5% NaOH-extracted straw	161.2	130.0	108.2	121.7
"Cellulose" from oat straw	119.5	101.5	81.8	76.9
Dextrose	91.5	70.5	59.7	64.3
Alfalfa	199.0	149.0	132.2	122.6
Soybeans 62/1 C:N	187.0	146.0	89.1	102.7
Soybeans 29/1 C:N	172.5	143.0	104.0	104.5

TABLE 16

Hypochlorite oxidation of straw during decomposition in absence of soil

(m.e. Cl per 100 gm. straw)

PLANT MATERIAL	RESIDUAL MATERIAL			ORIGINAL MATERIAL		
	After 112 days	After 322 days	After 596 days	After 112 days	After 322 days	After 596 days
Oat straw untreated	2670	3460	3067	1405	905	700
5% H_2SO_4-hydrolyzed straw	1750	2360	1616	1657	1666	1319
Cl-amine-extracted straw	1690	1800	1468	1555	1296	1000
0.5% Na_2CO_3-extracted straw	1850	2220	2027	1482	1297	1271
0.5% NaOH-extracted straw	1110	1830	3029	972	980	672

Norman (17) reported that there was an accumulation of lignin and nitrogenous complexes during aerobic decomposition processes, and that these were particularly susceptible to oxidation by hypochlorite. He found no clear evidence for a change in reactivity of the lignin during decomposition, but believed that the increase in reactivity might be taken as a measure of the accumulation of lignin and nitrogenous complexes. He suggested that this reagent might be used to study the residues from the same parent substance, but probably would not be suitable for comparison of residues of different sources and types, mainly because of the differences in reactivity and behavior of the lignin contained. The same general conclusions are reached from the work reported here.

Hypochlorite, because of the duration of the oxidation, would be less desirable than hypoiodite for studying the nature of organic matter. The activity of the latter reagent also seems to be more highly related to the lignin content than the former reagent, and for this reason, too, hypoiodite is of greater value as an aid in studying the nature of organic materials.

SUMMARY

Effects of composition of plant materials on properties of the decomposed residues have been studied by decomposing several plant materials of known composition, both in soil and in absence of soil, and analyzing the residues at several intervals over a period of 833 days, in order to follow as closely as possible the changes taking place during decomposition.

The composition of plant materials was ascertained to influence materially the quantity and the properies of the decomposed residues. Materials high in lignin were found to be much less decomposed than materials low in lignin, even after 28 months of decomposition.

The more active residues were also those derived from parent materials with a high lignin content. Oat straw cellulose and dextrose were found to decompose rapidly, and the residues from these materials were only slightly resistant to extraction by strong sulfuric acid in the cold, indicating that there was little, if any, lignified material in these residues; whereas there was considerable increase in this acid-resistant residue in soils receiving materials high in lignin.

The nitrogen content was maintained at a higher level in soils receiving the more resistant materials. This was probably due to the formation of lignoprotein, or some other complexes, because very little nitrogen was present in the form of ammonia or nitrate.

The fraction of the exchange capacity due to organic matter in these soils varied from 28 per cent in the check soil to 50 per cent in the soils receiving plant materials high in lignin. However, the increases in exchange capacities were not proportional to the increases in lignin content of the original materials, because of differences in the amount and nature of the decomposition and of the residues brought about by the pretreatments to which the straw was subjected.

The use of hypoiodite or hypochlorite as an aid in the study of the nature of soil organic matter was investigated. The activity of each reagent seems to be related to the lignin content; the latter agent also attacks the proteins.

Lignin seems to be more susceptible to oxidation by hypoiodite than by hypochlorite. For this reason, and because the oxidation by hypoiodite is complete in about 20 minutes, whereas the oxidation by hypochlorite may continue for days, the former reagent would be more suitable for studying the nature of soil organic matter. Alkaline hypoiodite may be helpful in detecting differences in the nature of organic matter in soils, as well as in the various layers of the same profile, particularly where the differences are likely to be due to the nature of the vegetation from which the organic matter was derived.

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